#### BIOSTATISTICS

# Probit Analysis: Assessing Goodness-of-Fit Based on Backtransformation and Residuals

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J. Econ. Entomol. 88(5): 1513-1516 (1995)

**ABSTRACT** We describe methods to convert complementary log-log-, logit-, probit-, log-complementary-log-log, log-logit, and log-probit-transformed data from bioassay experiments back to the original unit of measurement, the proportion of the test organisms responding to the tested stimulus; and for calculating residuals and standardized residuals (in the original units). We show how the method can be used to help select a model that best fits the bioassay data. We have developed a computer program for implementing the method.

KEY WORDS bioassay, probit analysis, transformation

PROBIT ANALYSIS IS used to analyze data from bioassay experiments, such as the proportions of insects killed by several concentrations of an insecticide or at several time intervals at 1 or more concentrations of an insecticide (Finney 1964). Results of probit analyses are reported typically as a concentration or time required to kill a certain proportion of the test insects (for example,  $LC_{50}$ ); the slope and intercept of the regression line of the probit-transformed data are also

reported (for example, Cilek and Greene 1994).

Goodness-of-fit of the regression line is indicated by

the chi-square.

Results of probit analyses are rarely reported in the original units, that is, proportion of insects killed. A researcher should examine and report the results in the original units because the purpose of a bioassay is to make inferences about the proportions of insects killed by the insecticide, not to make inferences about probits (Finney 1964). A plot of observed and predicted proportions of insects killed aids in assessing goodness-of-fit of the regression line. Goodness-of-fit also should be assessed by examination of residuals and standardized residuals in the original units, particularly to determine the possible causes of lack of fit when the

In addition to the probit transformation, the complementary log-log and logit transformations also are used to linearize bioassay data (Robertson and

chi-square is significant (Robertson and Preisler 1992).

Preisler 1992). Complementary log-log- and logit-transformed data are converted easily back to the original units. Converting probit-transformed data back to the original units is not straightforward; the conversion is most easily accomplished using tables (Beyer 1987) or mathematical computer programs (for example, Mathematica, Wolfram, Champaign, IL).

Here we show how data transformed back to proportion kill can be used to assess the goodness-of-fit of regression lines using data on the effects of malathion on *Bracon hebetor* Say (Hymenoptera: Braconidae), a parasitoid of insect pests of stored grain. We have developed a computer program that uses the slope and intercept from probit analysis programs (for example, Russell et al. 1977, Throne et al. 1995) to calculate the predicted proportion of insects killed by various concentrations of an insecticide or at various times by one concentration of an insecticide, including transforming the data from probits (or logits or the complementary log-log transformation) back to proportion killed. The program also can calculate residuals and standardized residuals.

## **Materials and Methods**

**Numerical Methods.** The predicted proportion of insects killed (*p*), in transformed units, is calculated as

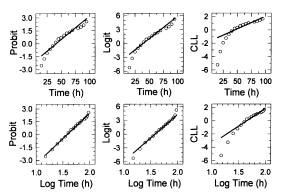
$$\hat{p} = a + bx, \tag{1}$$

where a = slope and b = intercept from the regression of transformed data and x is the concentration or time. If a logarithmic transformation of x was used in the original analysis, x is replaced by  $\log(x)$  in equation 1.

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**Fig. 1.** Observed (open circles) and predicted (line) probit, logit, complementary log-log, log-probit, log-logit, and log-complementary log-log transformations of proportion of the Savannah strain of *B. hebetor* killed over time by the LD<sub>99</sub> of malathion.

The formulas for calculating the predicted proportion of insects killed  $(\hat{q})$ , transformed back to the original units, are:

$$\hat{q} = 1 - e^{-e^{\hat{p}}} \tag{2}$$

for the complementary log-log transformation;

$$\hat{q} = \frac{e^{\hat{p}}}{1 + e^{\hat{p}}} \tag{3}$$

for the logit transformation; and

$$\hat{q} = \int_{-\pi}^{\hat{p}} \frac{e^{-u^2/2}}{\sqrt{2\pi}} du \tag{4}$$

for the probit transformation (Beyer 1987). Standardized residuals are calculated by dividing the residuals of proportion kill at each *x* by their standard errors (Snedecor and Cochran 1976):

$$SE = \sqrt{\frac{p(1-p)}{n}}, \tag{5}$$

where p = observed proportion killed and n number tested at each x. A computer program that transforms probit-type data back to the original units and calculates residuals and standardized residuals in the original units can be obtained from the first author.

**Example of Use.** We used the program to examine the goodness-of-fit of equations fit to data describing the time required to kill the Savannah strain of B. hebetor treated with the LD<sub>99</sub> of malathion determined for a field strain (Baker et al. 1995). The data were transformed before analysis with the complementary log-log, logit, or probit transformations of proportion kill and with and without a logarithmic transformation of x. Data subjected to a probit transformation of proportion of kill and a logarithmic transformation of x are referred to as log-probit transformed. We examined the fitted lines compared to both the transformed and untransformed observations, and examined residuals and standardized residuals of data converted back to the original units to determine goodness-of-fit.

#### Results

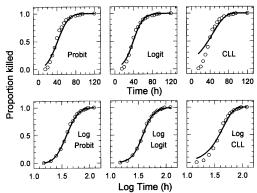
Chi-square values (26.11, 19.63, 72.20, 5.69, 6.28, and 20.09 for the probit, logit, complementary log-log, log-probit, log-logit, and log-complementary log-log transformations, respectively) and examination of the fitted regression lines compared with the transformed observations (Fig. 1) indicated that the log-probit and log-logit models had the best fit to the *B. hebetor* data. Residuals of transformed data (distance between observed points and fitted line) are not particularly helpful in determining which transformation results in the best fit because the residuals are not comparable for the 3 transformations because the scale varies with the transformation (that is, a residual of 1 for probittransformed data is not the same as a residual of 1 for logit-transformed data, when transformed back to proportion kill; Table 1). The probit corresponding to 10% kill is -1.282. Subtracting 1.0 from the probit (to attain a residual of -1) results in a probit of -2.282, which corresponds to 1.13% kill. Residuals of -1 on the logit and complementary log-log scales correspond with 3.93 and 3.80% kill.

For 90% kill, residuals of -1 on the probit, logit, and complementary log-log scales correspond to 61.09, 76.80, and 57.13% kill. These differences show that a residual of  $\pm$  1 is not the same for the 3 transformations when the data are transformed back to the original units

Table 1. Complementary log-log (CLL), logit, and probit transformations corresponding to selected levels of proportion of insects killed to demonstrate the effect of varying the transformed value by  $\pm 1$  at the 0. 10 and 0.90 levels of proportion of kill

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Proportion	Probit	Residual	Proportion	Logit	Residual	Proportion	CLL	Residual
0.10	-1.282		0.10	-2.197		0.10	-2.250	
0.01126	-2.282	0.08874	0.03927	-3.197	0.06073	0.03802	-3.250	0.06198
0.3891	-0.282	-0.2891	0.2320	-1.197	-0.1320	0.2490	-1.250	-0.1490
0.90	1.282		0.90	2.197		0.90	0.834	
0.6109	0.282	0.2891	0.7680	1.197	0.1320	0.5713	-0.166	0.3287
0.9887	2.282	-0.0887	0.9607	3.197	-0.0607	0.9981	1.834	-0.0981

The proportion kill corresponds with the transformed value that is  $\pm 1$  unit from the transformations for 0.10 and 0.90 kill. Residuals are either 0.10 or 0.90 kill minus the proportion kill corresponding to  $\pm 1$  transformation unit.



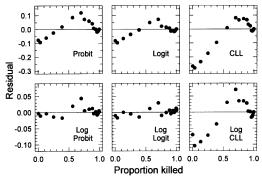
**Fig 2.** Observed (open circles) and predicted (line) proportion of the Savannah strain of B. hebetor killed over time by the LD<sub>99</sub> of malathion using the probit, logit, complementary log-log, log-probit, log-logit, and log-complementary log-log transformations for obtaining the prediction equations.

Thus, residuals of the transformed data are useful to determine whether the model fits the transformed data based on the presence of systematic trends in the pattern of the residuals, but are not useful for determining which of the transformations results in an equation which best describes the original data.

Residuals of data converted back to the original units are comparable among transformations and can be used to help determine which transformation best describes the original data. In our example, we noted a systematic trend to the residuals of probit-, logit-, complementary log-log-, and log-complementary log-log-transformed data. Equations developed by using these transformations overpredict the proportion kill early in the experiment.

We used output from the computer program to compare observed and predicted proportion of insects killed during each time interval (Fig. 2). The regression lines appear to fit the data better in the original units than in transformed units. For example, the predicted line fit the 1st and last data points in the log-logit plot in Fig. 1 poorly. However, when transformed back to the original units, the log-logit model fit all data points well (Fig. 2). Thus, the transformations may exaggerate the magnitude of the residuals of interest (deviation of observed and predicted proportion of kill). The plots of data in the original units directly show how well the prediction equations fit the observed data because the data--in the original unit of measurement, proportion kill--are reported in units that are easily understandable and comparable.

All 6 models appeared equally capable of describing the proportion kill at high values of proportion kill in this example, a conclusion confirmed by an examination of residuals calculated from data converted back to the original units (Fig. 3). The

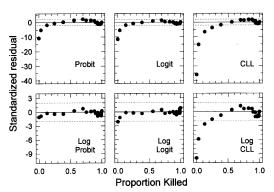


**Fig. 3.** Residual (observed minus predicted) proportion of the Savannah strain of B. hebetor killed by the  $LD_{99}$  of malathion versus the observed proportion killed using the probit, logit, complementary log-log, log-probit, log-logit, and log-complementary log-log transformations for obtaining the prediction equations.

log-probit and log-logit models fit the data best at lower values of proportion kill. Examination of standardized residuals confirms this result (Fig. 4), although only the log-probit transformation results in residuals that are all within  $\pm 2$  SD of zero. Preisler (1988) suggested that standardized residuals lying more than  $\pm 2$  SD from zero indicate possible lack of fit.

# Discussion

Results from probit analyses are rarely reported in the original units (proportion of insects killed), a practice that makes it difficult to assess the actual practicality of the regression equation obtained from the analysis. Our program provides an easy method for converting the results of probit analyses back to the original units, and the resulting data can be used to plot observed and predicted proportion of insects killed as in Fig. 2.



**Fig. 4.** Standardized residual proportion of the Savannah strain of *B. hebetor* killed by the LD<sub>99</sub> of malathion versus the observed proportion killed using the probit, logit, complementary log-log, log-probit, log-logit, and log-complementary log-log transformations for obtaining the prediction equations.

Plotting permits assessment of the regression equation in units that are easily understandable. Examination of plots of residuals and standardized residuals in the original units aids in assessing goodness of fit.

Naylor (1964) reported that the choice of the complementary log-log, logit, or probit transformations had little effect on goodness-of-fit to several biological data sets. However, our data indicate that, for insect bioassay data, the choice of transformation may have a great effect on goodness-of-fit, as indicated by chisquare analysis and plots of the data and residuals.

Converting transformed data back to the original units is particularly important in selecting which transformation results in a model that best fits the data. As demonstrated here, comparison of transformed residuals to determine which model fits the data best is not valid because the residuals are on different scales. The data must be transformed back to the original units (proportion kill) so that all residuals are on the same scale and can be compared. In addition to an examination of chi-square values, plots of observed versus predicted proportion of kill and residuals in the original units, variance estimates should also be considered in selecting the model which best fits the data. If the intent of the bioassay is to determine the concentration or time that is required to kill a certain proportion of the insects, then the researcher will want to choose a model that also minimizes the confidence limits on that lethal concentration or time value.

## Acknowledgments

We thank Walter P. Gould (USDA-ARS, Miami, FL) and Bruce E. Mackey (USDA-ARS, Albany, CA) for reviewing an earlier version of the manuscript, and Victor Chew (USDA-ARS, Gainesville, FL) for statistical advice.

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Received for publication 14 September 1994; accepted 22 May 1995.